

# Anti-ECHS1 Antibody Picoband™

Catalog Number: A04508-1

#### **About ECHS1**

Enoyl Coenzyme A hydratase, short chain, 1, mitochondrial, also known as ECHS1, is a human gene. The protein encoded by this gene functions in the second step of the mitochondrial fatty acid beta-oxidation pathway. It catalyzes the hydration of 2-trans-enoyl-coenzyme A (CoA) intermediates to L-3-hydroxyacyl-CoAs. The gene product is a member of the hydratase/isomerase superfamily. It localizes to the mitochondrial matrix. Transcript variants utilizing alternative transcription initiation sites have been described in the literature.

#### Overview

Product Name	Anti-ECHS1 Antibody Picoband™
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-ECHS1 Antibody Picoband™ catalog # A04508-1. Tested in ELISA, Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Mouse, Rat.
Application	ELISA, Flow Cytometry, IF, IHC, ICC, WB
Clonality	Polyclonal
Formulation	Each vial contains 4 mg Trehalose, 0.9 mg NaCl, 0.2 mg Na2HPO4.
Storage Instructions	At -20°C for one year from date of receipt. After reconstitution, at 4°C for one month. It can also be aliquotted and stored frozen at -20°C for six months. Avoid repeated freezing and thawing.
Host	Rabbit
Uniprot ID	P30084

### **Technical Details**

Immunogen	E.coli-derived human ECHS1 recombinant protein (Position: R12-Q290).
Recommended Detection Systems	Boster recommends Enhanced Chemiluminescent Kit with anti-Rabbit IgG (EK1002) for Western blot, and HRP Conjugated anti-Rabbit IgG Super Vision Assay Kit (SV0002-1) for IHC(P) and ICC.
Cross Reactivity	No cross-reactivity with other proteins.
Isotype	Rabbit IgG
Form	Lyophilized
Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 ug/ml.
Purification	Immunogen affinity purified.
Suggested Dilutions	Dilute the sample so that the expected range of concentrations fall within the detection range of this



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kit.
If the expected range of concentration is unknown, a pilot test should be conducted to decide the optimal dilution ratio for your samples.
Some PubMed article(s) citing the expression level of this target are as follows:
Boster Bio's internal QC testing used:
Western blot, 0.25-0.5 ug/ml, Human, Mouse, Rat
Immunohistochemistry(Paraffin-embedded Section), 2-5 ug/ml, Human, Rat
Immunocytochemistry/Immunofluorescence, 5 ug/ml, Human
Immunofluorescence, 5 ug/ml, Human
Flow Cytometry, 1-3 ug/1x10^6 cells, Human
Direct ELISA, 0.1-0.5 ug/ml, Human



## Anti-ECHS1 Antibody Picoband™ (A04508-1) Images

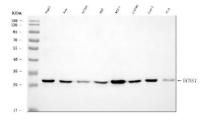


Figure 1. Western blot analysis of ECHS1 using anti-ECHS1 antibody (A04508-1).

Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions.

Lane 1: human HepG2 whole cell lysates,

Lane 2: human Hela whole cell lysates,

Lane 3: human HT1080 whole cell lysates,

Lane 4: human 293T whole cell lysates,

Lane 5: human MCF-7 whole cell lysates,

Lane 6: human U-87MG whole cell lysates,

Lane 7: human CACO-2 whole cell lysates,

Lane 8: human PC-3 whole cell lysates.

After electrophoresis, proteins were transferred to a nitrocellulose membrane at 150 mA for 50-90 minutes. Blocked the membrane with 5% non-fat milk/TBS for 1.5 hour at RT. The membrane was incubated with rabbit anti-ECHS1 antigen affinity purified polyclonal antibody (Catalog # A04508-1) at 0.5 ug/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody at a dilution of 1:5000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1002) with Tanon 5200 system. A specific band was detected for ECHS1 at approximately 29 kDa. The expected band size for ECHS1 is at 29 kDa.

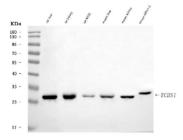


Figure 2. Western blot analysis of ECHS1 using anti-ECHS1 antibody (A04508-1).

Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions.

Lane 1: rat liver tissue lysates,

Lane 2: rat kidney tissue lysates,

Lane 3: rat RH35 whole cell lysates,

Lane 4: mouse liver tissue lysates,

Lane 5: mouse kidney tissue lysates,

Lane 6: mouse HEPA1-6 whole cell lysates.

After electrophoresis, proteins were transferred to a nitrocellulose membrane at 150 mA for 50-90 minutes. Blocked the membrane with 5% non-fat milk/TBS for 1.5 hour at RT. The membrane was incubated with rabbit anti-ECHS1 antigen affinity purified polyclonal antibody (Catalog # A04508-1) at 0.5 ug/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody at a dilution of 1:5000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1002) with Tanon 5200 system. A specific band was detected for ECHS1 at approximately 29 kDa. The expected band size for ECHS1 is at 29 kDa.



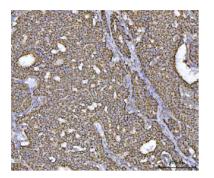


Figure 3. IHC analysis of ECHS1 using anti-ECHS1 antibody (A04508-1).

ECHS1 was detected in a paraffin-embedded section of human breast cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-ECHS1 Antibody (A04508-1) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.

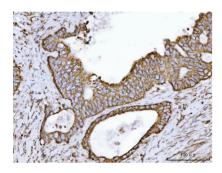


Figure 4. IHC analysis of ECHS1 using anti-ECHS1 antibody (A04508-1).

ECHS1 was detected in a paraffin-embedded section of human colorectal adenocarcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-ECHS1 Antibody (A04508-1) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.

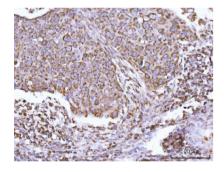


Figure 5. IHC analysis of ECHS1 using anti-ECHS1 antibody (A04508-1).

ECHS1 was detected in a paraffin-embedded section of human esophageal squamous carcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-ECHS1 Antibody (A04508-1) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.

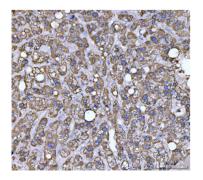


Figure 6. IHC analysis of ECHS1 using anti-ECHS1 antibody (A04508-1).

ECHS1 was detected in a paraffin-embedded section of human liver cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-ECHS1 Antibody (A04508-1) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.



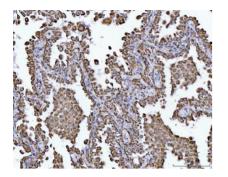


Figure 7. IHC analysis of ECHS1 using anti-ECHS1 antibody (A04508-1).

ECHS1 was detected in a paraffin-embedded section of human lung cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-ECHS1 Antibody (A04508-1) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.

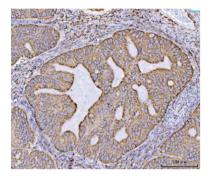


Figure 8. IHC analysis of ECHS1 using anti-ECHS1 antibody (A04508-1).

ECHS1 was detected in a paraffin-embedded section of human ovarian cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-ECHS1 Antibody (A04508-1) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.

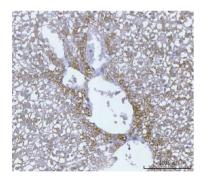


Figure 9. IHC analysis of ECHS1 using anti-ECHS1 antibody (A04508-1).

ECHS1 was detected in a paraffin-embedded section of rat liver tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-ECHS1 Antibody (A04508-1) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.

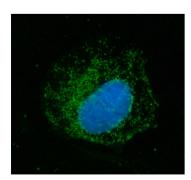
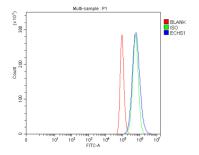
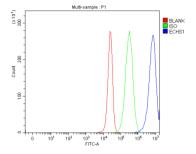


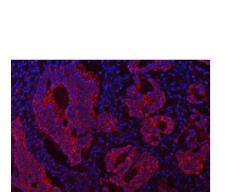
Figure 10. IF analysis of ECHS1 using anti-ECHS1 antibody (A04508-1).

ECHS1 was detected in an immunocytochemical section of CACO-2 cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5 ug/mL rabbit anti-ECHS1 Antibody (A04508-1) overnight at 4°C. DyLight® 488 Conjugated Goat Anti-Rabbit IgG (BA1127) was used as secondary antibody at 1:100 dilution and incubated for 30 minutes at 37°C. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.









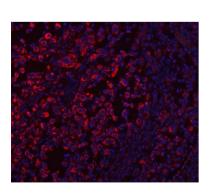


Figure 11. Flow Cytometry analysis of Hela cells using anti-ECHS1 antibody (A04508-1).

Overlay histogram showing Hela cells stained with A04508-1 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-ECHS1 Antibody (A04508-1, 1 ug/1x $10^6$  cells) for 30 min at 20°C. DyLight® 488 conjugated goat anti-rabbit IgG (BA1127, 5-10 ug/1x $10^6$  cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1 ug/1x $10^6$ ) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

Figure 12. Flow Cytometry analysis of 293T cells using anti-ECHS1 antibody (A04508-1).

Overlay histogram showing 293T cells stained with A04508-1 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-ECHS1 Antibody (A04508-1, 1 ug/1x10 $^6$  cells) for 30 min at 20 $^\circ$ C. DyLight® 488 conjugated goat anti-rabbit IgG (BA1127, 5-10 ug/1x10 $^6$  cells) was used as secondary antibody for 30 minutes at 20 $^\circ$ C. Isotype control antibody (Green line) was rabbit IgG (1 ug/1x10 $^6$ ) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

Figure 13. IF analysis of ECHS1 using anti-ECHS1 antibody (A04508-1).

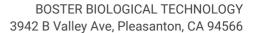
ECHS1 was detected in a paraffin-embedded section of human intestinal cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 5 ug/mL rabbit anti-ECHS1 Antibody (A04508-1) overnight at 4°C. DyLight®550 Conjugated Goat Anti-Rabbit IgG (BA1135) was used as secondary antibody at 1:500 dilution and incubated for 30 minutes at 37°C. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.

Figure 14. IF analysis of ECHS1 using anti-ECHS1 antibody (A04508-1).

ECHS1 was detected in a paraffin-embedded section of human lung cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 5 ug/mL rabbit anti-ECHS1 Antibody (A04508-1) overnight at 4°C. DyLight® 550 Conjugated Goat Anti-Rabbit IgG (BA1135) was used as secondary antibody at 1:500 dilution and incubated for 30 minutes at 37°C. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.











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Anti-ECHS1 Antibody